CLAIMS

What is claimed is:

1. A protein microarray, comprising:
   a solid support;
   a linker covalently attached to said solid support; and
   a protein or protein fragment having a terminus that is capable of forming a covalent bond with said linker.

2. The microarray of claim 1, wherein said terminus is a carboxy terminus.

3. The microarray of claim 1, wherein said solid support is glass.

4. The microarray of claim 1, wherein said linker comprises a maleimide group.

5. The microarray of claim 1, wherein said linker comprises a vinyl sulfone group.

6. The microarray of claim 1, wherein said linker comprises a N-hydroxy succinimide group.

7. The microarray of claim 1, wherein said protein or protein fragment is an antibody or antibody fragment.

8. The microarray of claim 7, wherein said antibody or antibody fragment is a single chain antibody.

9. The microarray of claim 1, wherein said microarray has at least 1,000 spots per cm^2.

10. The microarray of claim 1, wherein said microarray has at least 2,000 spots per cm^2.

11. A method for attaching a protein to a support surface, said method comprising the steps of:

(a) covalently attaching a bovine serum albumin molecule to a support surface;

(b) forming an activated carbamate group or activated ester group on an exposed surface of said molecule; and

(c) exposing said activated carbamate group or said activated ester group to a binding element comprising an amine, thereby forming a covalent bond between said carbamate or said ester group of said molecule and said amine group of said binding element.

12. The method of claim 11, wherein said forming step comprises exposing said bovine serum albumin to a reagent to form a N-hydroxy succinimide group.

13. The method of claim 11, wherein said binding element is a protein.

14. The method of claim 13, wherein said protein is an antibody or antibody fragment.

15. The method of claim 14, wherein said antibody or antibody fragment is a single chain antibody.

16. The method of claim 11, further comprising the step of blocking any of said activated carbamate or ester groups that have not bound to said binding element.

17. A method for attaching a protein to a support surface, said method comprising the steps of:

(a) providing a support surface comprising a first chemical group available for reaction;

(b) providing a capture protein comprising a first terminus and a second terminus, said first terminus capable of binding to a ligand, said second terminus comprising a second chemical group; and

(c) forming a covalent bond between said first chemical group and said second chemical group, thereby attaching said capture protein to said support surface at said second terminus of said capture protein.

18. The method of claim 17, wherein said capture protein comprises a terminal cysteine.

19. The method of claim 18, wherein said terminal cysteine is at a carboxy terminal.

20. The method of claim 18, wherein said forming step comprises chemically reducing said cysteine.

21. A method for identifying a small molecule regulator of protein binding, the method comprising the steps of:

(a) attaching a capture protein on a support surface;

(b) exposing said substrate to a ligand for said capture protein and at least one small molecule; and

(c) detecting the presence or the absence of binding between said capture protein and said ligand.

22. The method of claim 21, wherein step (a) comprises attaching said capture protein on a BSA-NHS slide.

23. The method of claim 21, wherein step (a) comprises functionalizing said support surface with aldehyde groups.

24. The method of claim 21, wherein step (a) comprises attaching said capture protein in a microarray of at least 1,000 spots per cm^2.

25. The method of claim 21, further comprising fusing said capture protein to a GST protein.

26. The method of claim 21, further comprising detecting said binding between said capture protein and said ligand through a fluorescent dye.

27. The method of claim 26, wherein said fluorescent dye comprises a hydrophilic polymer moiety.

28. The method of claim 27, wherein said moiety is a polyethyleneglycol.

29. The method of claim 21, wherein step (c) comprises detecting said binding between said capture protein and said ligand through a labeled phage particle displaying an antibody fragment.

30. The method of claim 21, wherein said ligand comprises a family of related proteins.

31. The method of claim 30, wherein said ligand comprises the Bcl-2 family of proteins.

32. The method of claim 21, wherein said capture protein comprises a family of related proteins.

33. A method for identifying a small molecule that selectively affects a cellular pathway, the method comprising the steps of:

(a) attaching a microarray of capture proteins on a support surface, said microarray comprises proteins that act in a cellular pathway;

(b) exposing said substrate surface to at least one ligand of said capture proteins and at least one small molecule; and

(c) detecting a change in binding between said capture proteins and said ligand, said change resulting from interaction with said small molecule.

34. The method of claim 33, wherein step (c) further comprises using mass spectrometry to quantify said change.

35. The method of claim 33, further comprising detecting said binding between said capture protein and said ligand through a fluorescent dye.

36. The method of claim 35, wherein said fluorescent dye comprises a hydrophilic polymer moiety.

37. The method of claim 36, wherein said moiety is a polyethyleneglycol.

38. The method of claim 33, wherein step (c) comprises detecting said binding between said capture protein and said ligand through a labeled phage particle displaying an antibody fragment.

39. The method of claim 33, wherein step (a) comprises attaching said capture proteins on a BSA-NHS slide.

40. The method of claim 34, wherein step (a) comprises attaching said capture protein in a microarray of at least 1,000 spots per cm^2.

41. A method for labeling an antigen, said method comprising:

digesting an antigen with a protease thereby to produce multiple peptides such that at least one of said peptides is capable of receiving a label at a region of said peptide that does not interfere with binding between an epitope on said peptide and an antibody or antibody fragment.

42. The method of claim 41, further comprising using a succinimidyl ester dye to label said peptide.

43. The method of claim 42, wherein said succinimidyl ester dye is Cy3, Cy5 or an Alexa dye.

44. The method of claim 41, further comprising labeling only a terminal primary amine of said peptide, wherein said epitope is internal.

45. The method of claim 41, further comprising digesting said antigen with trypsin.

46. A method for detecting a phorsphorylated protein, the method comprising the steps of:

(a) fragmenting a candidate protein into a plurality of peptides comprising a target peptide, the target peptide comprising a phorsphorylation site;

(b) exposing said plurality of peptides to an antibody or antibody fragment having affinity for an epitope on said target peptide adjacent to said phorsphorylation site;

(c) selecting said target peptide based on affinity of said target peptide for said antibody or antibody fragment; and

(d) conducting mass spectrometry on said target peptide to detect the presence of a subset of said protein that has been phorsphorylated.

47. The method of claim 46 wherein step (a) comprises digesting said candidate protein with a protease.

48. The method of claim 47, wherein the protease is trypsin.

49. The method of claim 46 further comprising panning an scFv against said epitope.

50. The method of claim 46 wherein step (c) comprises immobilizing said antibody or antibody fragment to a solid support.

51. The method of claim 46 wherein step (d) comprises detecting a change in the molecular weight of a subset of said target peptide.

52. The method of claim 46 wherein step (d) comprises conducting MALDI mass spectrometry.

53. The method of claim 46, further comprising immunizing a monoclonal antibody against the epitope.

54. The method of claim 46, further comprising immunizing a polyclonal antibody against the epitope.

55. The method of claim 46 wherein the epitope is less than 15 amino acids away from the phorsphorylation site.

56. The method of claim 46 wherein the epitope is less than 10 amino acids away from the phorsphorylation site.

57. The method of claim 46 wherein the epitope is less than 10 amino acids.

58. The method of claim 46 wherein the epitope is less than 5 amino acids

59. A method of studying a cellular event, the method comprising the steps of:

(a) attaching a capture molecule on a support surface, said capture molecule having affinity for a ligand;

(b) exposing said substrate surface to a solution containing a cellular organelle, said ligand associated with a surface of said organelle; and

(c) capturing said organelle through binding between said capture molecule and said ligand.

60. The method of claim 59, wherein said capture molecule comprises a protein.

61. The method of claim 59, wherein said capture molecule comprises an antibody or a fragment thereof.

62. The method of claim 59, further comprising studying a protein associated with said captured organelle.

63. The method of claim 59, wherein said organelle is a mitochondria.

64. The method of claim 63, wherein said ligand is a voltage dependent anion channel receptor that is uniquely associated with the mitochondria membrane.

65. The method of claim 59 wherein said solution is a whole-cell extract.

66. The method of claim 59 wherein said solution is a fraction of a whole-cell extract.

67. The method of claim 59, further comprising detecting said capturing through a fluorescent dye.

68. The method of claim 67, wherein said fluorescent dye comprises a hydrophilic polymer moiety.

69. The method of claim 68, wherein said moiety is a polyethyleneglycol.

70. The method of claim 67 wherein the dye has potentiometric quality for recognizing intact voltage gradient of said organelle.

71. The method of claim 70 wherein said organelle is a mitochondria.

72. The method of claim 59, further comprising detecting said capturing through a labeled phage particle displaying an antibody fragment.